

I CLAIM:

1. A purified and isolated nucleic acid having a nucleotide sequence encoding an A chain of a ricin-like toxin, a B chain of a ricin-like toxin and a heterologous linker amino acid sequence linking the A and B chains, the
5 heterologous linker sequence containing a cleavage recognition site for a protease localized in cells or tissues affected by a specific disease.
2. The nucleic acid sequence of claim 1 wherein the linker sequence encodes a peptide wherein at least 5 amino acids differ from the wild-type.
3. The nucleic acid sequence of claim 2 wherein the linker sequence
10 contains a cleavage recognition site recognized by a protease selected from the group consisting of: a cancer associated protease, a viral protease, a fungal protease, and a parasite protease.
4. A nucleic acid sequence of claim 3 wherein the A chain is ricin A chain, abrin toxin A chain, diphtheria toxin A chain, or Domain I of
15 Pseudomonas exotoxin.
5. A nucleic acid molecule of claim 3 wherein the A chain is volkensin toxin A chain, cholera toxin A chain, modeccin toxin A chain, viscumin toxin A chain or shiga toxin A chain.
6. A nucleic acid sequence of claim 3 wherein the B chain is ricin B
20 chain, abrin toxin A chain, diphtheria toxin B chain, or Domain II/III of Pseudomonas exotoxin.
7. A nucleic acid molecule of claim 3 wherein the B chain is volkensin toxin B chain, cholera toxin B chain, modeccin toxin B chain, viscumin toxin B chain or shiga toxin B chain.
- 25 8. A nucleic acid sequence of claim 3 wherein the cleavage recognition site is recognized by a cancer-associated protease which is selected from

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the group consisting of: cathepsin B, a matrix metalloproteinase, cathepsin L, cathepsin D, urokinase-type plasminogen activator, tissue-type plasminogen activator, human prostate-specific antigen, kallikrein, neutrophil elastase, and calpain.

- 5 9. A nucleic acid sequence of claim 3 wherein the cleavage recognition site is recognized by a parasitic protease which is a *Plasmodium falciparum* protease.
10. A nucleic acid sequence of claim 3 wherein the cleavage recognition site is recognized by viral protease which is selected from the group consisting of: human cytomegalovirus, human herpes virus, varicella zoster virus, hepatitis A virus, hepatitis C virus, Epstein-Barr virus-specific protease, and infectious laryngotracheitis virus.
- 10 11. A nucleic acid sequence of claim 3 wherein the cleavage recognition site is recognized by fungal protease which is a *Candida* acid protease.
- 15 12. A nucleic acid sequence of claim 3 having the nucleotide sequence according to SEQ ID No. 3; SEQ ID No 5; SEQ ID No 7; SEQ ID No 9; SEQ ID No 11; SEQ ID No 13; SEQ ID No 15; SEQ ID No 17; SEQ ID No 19; SEQ ID No 21; SEQ ID No 23; SEQ ID No 25; SEQ ID No 27; SEQ ID No 29; SEQ ID No 31; SEQ ID No 33; SEQ ID No 35; SEQ ID No 37; SEQ ID No 39; SEQ ID No 48; SEQ ID No 50; SEQ ID No 52; SEQ ID No 54; SEQ ID No 74; SEQ ID No 77; SEQ ID No 80; SEQ ID No 83; SEQ ID No 86; SEQ ID No 89; SEQ ID No 92; SEQ ID No 95; SEQ ID No 98; SEQ ID No 101; SEQ ID No 104; SEQ ID No 107; SEQ ID No 110; SEQ ID No 113; SEQ ID No 116; SEQ ID No 119; SEQ ID No 122; or SEQ ID No 125.
- 20 13. A plasmid incorporating the nucleic acid of claim 12.
- 25 14. A baculovirus transfer vector incorporating the nucleic acid of claim 12.

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15. A recombinant protein comprising an A chain of a ricin-like toxin, a B chain of a ricin-like toxin and a heterologous linker amino acid sequence, linking the A and B chains, wherein the linker sequence contains a cleavage recognition site for a protease localized in cells or tissues affected by a specific disease.
16. The recombinant protein of claim 15 wherein at least 5 amino acids of the linker amino acid sequence differ from the wild-type.
17. The recombinant protein of claim 16 wherein the linker sequence contains a cleavage recognition site which is recognized by a protease selected from the group consisting of: a cancer, viral, fungal, and a parasitic protease.
18. A recombinant protein of claim 17 wherein the A chain is ricin A chain, abrin toxin B chain, diphtheria toxin A chain, or Domain I of Pseudomonas exotoxin.
19. A recombinant protein of claim 17 wherein the A chain is volkensin toxin A chain, cholera toxin A chain, modeccin toxin A chain, viscumin toxin A chain, or shiga toxin A chain.
20. A recombinant protein of claim 17 wherein the B chain is ricin B chain, abrin toxin B chain, diphtheria toxin B chain, or Domain II/III of Pseudomonas exotoxin.
21. A recombinant protein of claim 17 wherein the B chain is volkensin toxin B chain, cholera toxin B chain, modeccin toxin B chain, viscumin toxin B chain, or shiga toxin B chain.
22. A recombinant protein of claim 17 wherein the cleavage recognition site is recognized by a cancer-associated protease selected from the group consisting of: cathepsin B, a matrix metalloproteinase, cathepsin L, cathepsin D, urokinase-type plasminogen activator, tissue-type

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plasminogen activator, human prostate-specific antigen, kallikrein, neutrophil elastase, and calpain.

23. A recombinant protein of claim 17 wherein the cleavage recognition site is recognized by a parasitic protease which is a
5 Plasmodium falciparum protease.

24. A recombinant protein of claim 17 wherein the cleavage recognition site is recognized by a viral protease which is selected from the group consisting of: human cytomegalovirus, human herpes virus, varicella zoster virus, hepatitis A virus, hepatitis C virus, Epstein-Barr
10 virus-specific protease, and infectious laryngotracheitis virus.

25. A recombinant protein of claim 17 wherein the cleavage recognition site is recognized by a fungal protease which is a Candida acid protease.

26. A recombinant protein of claim 17 having the linker amino acid
15 sequence according to SEQ ID No. 40; SEQ ID No. 41; SEQ ID No. 42; SEQ ID No. 43; SEQ ID No. 44; SEQ ID No. 45; SEQ ID No. 46; SEQ ID No. 55; SEQ ID No. 56; SEQ ID No. 57; SEQ ID No. 58; SEQ ID No. 59; SEQ ID No. 60; SEQ ID No. 61; SEQ ID No. 62; SEQ ID No. 63; SEQ ID No. 64; SEQ ID No. 65; SEQ ID No. 66; SEQ ID No. 67; SEQ ID No. 68; SEQ ID No. 69; SEQ
20 ID No. 70; SEQ ID No. 71; SEQ ID No. 72; SEQ ID No. 75; SEQ ID No. 78; SEQ ID No. 81; SEQ ID No. 84; SEQ ID No. 87; SEQ ID No. 90; SEQ ID No. 93; SEQ ID No. 96; SEQ ID No. 99; SEQ ID No. 102; SEQ ID No. 105; SEQ ID No. 108; SEQ ID No. 111; SEQ ID No. 114; SEQ ID No. 117; SEQ ID No. 120; SEQ ID No. 123; or SEQ ID No. 126.

25 27. A method of inhibiting or destroying cells of a tissue affected by a disease, wherein a protease specific to the disease is localized in the cells or tissue comprising the steps of:

(a) preparing a purified and isolated nucleic acid having a nucleotide sequence encoding an A chain of a ricin-like toxin, a B chain of a

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ricin-like toxin, and a heterologous linker amino acid sequence, linking the A and B chains, wherein the linker sequence contains a cleavage recognition site for the protease;

5 (b) introducing the nucleic acid into a host cell and expressing the nucleic acid in the host cell to obtain a recombinant protein comprising an A chain of a ricin-like toxin, a B chain of a ricin-like toxin and a linker amino acid sequence;

(c) suspending the protein in a pharmaceutically acceptable carrier, diluent or excipient, and

10 (d) contacting the cells with the recombinant protein.

28. A method according to claim 27 wherein the linker sequence encodes a peptide wherein at least 5 amino acids differ from the wild-type.

29. The method of claim 28 where the disease is one of cancer, a fungal infection, or cells infected with a virus or parasite.

15 30. A method of inhibiting or destroying cells of a tissue affected by a disease, wherein a protease specific to the disease is localized in the cells or tissue comprising the step of contacting the cells with a recombinant protein according to claim 26.

20 31. A method of treating a disease comprising administering a recombinant protein according to claim 26 to an animal in need thereof.

32. A method of treating a disease comprising administering a nucleic acid molecule according to claim 12 to an animal in need thereof.

25 33. A method of treating a mammal with cancer or infected with a fungus, virus or parasite, comprising the steps of preparing a recombinant protein of claim 15 wherein the linker sequence contains a cleavage recognition site for a cancer, fungal, viral or parasitic protease and administering the protein to the mammal.

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34. A method of inhibiting or destroying cells affected by a disease associated with a protease, comprising contacting cells affected by a disease associated with the protease with a recombinant ricin protein such that cells affected by a disease associated with a protease are inhibited or
5 destroyed, wherein said recombinant ricin protein has an A chain of a ricin-like toxin, B-chain of a ricin-like toxin, and a heterologous linker amino acid sequence containing a cleavage recognition site for the protease associated with the disease.

35. A method according to claim 33, wherein said heterologous linker
10 amino acid sequence contains at least five amino acids which differ from the wild-type.

36. The method of claim 34 where the disease is one of cancer, a fungal infection, or cells infected with a virus or parasite.

37. A process for preparing a pharmaceutical for treating a mammal
15 with cancer, fungal infection, viral infection or parasitic infection, comprising the steps of :

(a) preparing a purified and isolated nucleic acid having a nucleotide sequence encoding an A chain of a ricin-like toxin, a B chain of a ricin-like toxin, and a heterologous linker amino acid sequence, linking the
20 A and B chains, wherein the linker sequence contains a cleavage recognition site for a cancer, viral or parasitic protease;

(b) introducing the nucleic acid into a host cell and expressing the nucleic acid in the host cell to obtain a recombinant protein comprising an A chain of a ricin-like toxin, a B chain of a ricin-like toxin and a linker amino
25 acid sequence;

(c) suspending the protein in a pharmaceutically acceptable carrier, diluent or excipient.

38. A pharmaceutical composition for treating cancer or a fungal, or viral, or parasitic infection in an animal comprising the recombinant

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protein of claim 16 and a pharmaceutically acceptable carrier, diluent or excipient.

39. A pharmaceutical composition for treating cancer or a fungal, or viral, or parasitic infection in an animal comprising the nucleic acid molecule of claim 3 and a pharmaceutically acceptable carrier, diluent or excipient.
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